

(19) World Intellectual Property
Organization
International Bureau



(43) International Publication Date
27 May 2004 (27.05.2004)

PCT

(10) International Publication Number
WO 2004/044132 A3

(51) International Patent Classification⁷: C12P 19/34,
A01N 43/04, C07H 21/00, 21/02, 21/04

(74) Agents: CALDWELL, John, W. et al.; Woodcock Washburn LLP, One Liberty Place - 46th Floor, Philadelphia, PA 19103 (US).

(21) International Application Number:
PCT/US2003/035063

(81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KG, KF, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(22) International Filing Date:
4 November 2003 (04.11.2003)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
60/423,760 5 November 2002 (05.11.2002) US

(84) Designated States (regional): ARIPO patent (BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

(71) Applicant (for all designated States except US): ISIS PHARMACEUTICALS, INC. [US/US]; 2292 Faraday Avenue, Carlsbad, CA 92008 (US).

(72) Inventors; and

Published:

— with international search report
— before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments

(75) Inventors/Applicants (for US only): BAKER, Brenda, F. [US/US]; 2147 Avenida Toronja, Carlsbad, CA 92009 (US). ELDRUP, Anne, B. [DK/US]; 1190 Encinitas Boulevard, # 252, Encinitas, CA 92024 (US). MANOHARAN, Muthiah [US/US]; 25 Circle Drive, Weston, MA 02493 (US). BHAT, Balkrishen [IN/US]; 911 Rosemary Avenue, Carlsbad, CA 92009 (US). GRIFFEY, Richard [US/US]; 360 Barsby Street, Vista, CA 92084 (US). SWAYZE, Eric, E. [US/US]; 7789 Palenque Street, Carlsbad, CA 92009 (US). CROOKE, Stanley, T. [US/US]; 3211 Piragua Street, Carlsbad, CA 92009 (US).

(88) Date of publication of the international search report:
7 October 2004

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: MODIFIED OLIGONUCLEOTIDES FOR USE IN RNA INTERFERENCE

(57) Abstract: The present invention provides modified oligonucleotides for use in the RNA interference pathway of gene modulation. The modified oligonucleotides are also provided having a 3' terminal cap group.

WO 2004/044132 A3

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US03/35063

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12P 19/34; A01N 43/04; C07H 21/00, 21/02, 21/04
 US CL : 435/91.31; 514/44, 536/23.1, 24.1, 24.5, 25.3

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 435/91.31; 514/44, 536/23.1, 24.1, 24.5, 25.3

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
 Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category * | Citation of document, with indication, where appropriate, of the relevant passages | Relevant to claim No. |
|------------|--|-----------------------------------|
| P, X | US 2003/0143732 A1 (FOSNAUGH et al.) 31 July 2003 (31.7.2003) pgs. 3-11, 19-23 in particular | 1-40 |
| P, X | CZAUVERNA et al. Structural variations and stabilising modifications of synthetic siRNAs in mammalian cells. Nucleic Acids Research. June 2003. Vol. 31., pp. 2705-2716 (entire document) | 1-17, 32, 35 |
| Y | | 18-31, 38-40 |
| Y | PADDISON et al. Stable suppression of gene expression by RNAi in mammalian cells. Proc. Natl. Acad. Sci. February 2002. Vol. 99. pp. 1443-1448 (entire document) | 1-40 |
| Y | SUI et al. A DNA vector -based RNAi technology to suppress gene expression in mammalian cells. Proc. Natl. Acad. Sci. April 2002. Vol. 99. pp. 5515-5520 (entire document) | 1-17, 26-30, 32, 35, 38 |
| Y | ELBASHIR et al. Duplexes of 21-nucleotide RNAs mediate RNA interference in cultured mammalian cells. Nature May 2001. Vol. 411. pp. 494-498 (entire document) | 1-17, 26-30, 32, 34-45, 37-38, 40 |
| Y | CAPLEN et al. Specific inhibition of gene expression by small double-stranded RNAs in invertebrate and vertebrate systems. Proc. Natl. Acad. Sci. August 2001. Vol. 98. pp. 9742-47 (entire document). | 1-25, 32-33, 35-36, 38-39 |

☒ Further documents are listed in the continuation of Box C.☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"B" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority date(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to oral disclosures, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and set in conflict with the application but cited to understand the principle, or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"Z" document member of the same patent family

Date of the actual completion of the international search

28 June 2004 (28.06.2004)

Date of mailing of the international search report

28 AUG 2004

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US
 Commissioner for Patents
 P.O. Box 1459
 Alexandria, Virginia 22313-1450
 Facsimile No. (703) 305-3230

Authorized officer

Jon B. Ashen

Telephone No. 703-308-1235

Form PCT/ISA/210 (second sheet) (July 1998)

INTERNATIONAL SEARCH REPORT

PCT/US03/35063

C. (Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

| Category * | Citation of document, with indication, where appropriate, of the relevant passages | Relevant to claim No. |
|------------|---|-----------------------|
| Y | YU et al. RNA interference by expression of short-interfering RNAs and hairpin RNAs in mammalian cells. Proc. Natl. Acad. Sci. April 2002. Vol. 99 pp. 6047-6052 (entire document). | 1-40 |
| Y | BEIGELMAN et al. Chemical modification of hammerhead ribozymes. Catalytic activity and nuclease resistance. J. Biol. Chem. October 1995. Vol. 43. pp. (entire document) | 1-40 |

INTERNATIONAL SEARCH REPORT

PCT/US03/35063

Continuation of B. FIELDS SEARCHED Item 3:

STN/CAS

Search terms: RNAi, siRNA, shRNA, RNAseIII, DICER, RNA-induced silencing complex, RISC, 3'-terminal cap, 3'-5' linkage, antisense oligomer/oligonucleotide, ribozyme, abasic nucleoside, abasic nucleotide